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42. The system of claim 39, wherein the thermal cyclers is computer-controlled.---

REMARKS

Reconsideration and withdrawal of the rejections set forth in the Office action dated August 11, 1999 are respectfully requested. The applicant petitions the Commissioner for a three-month extension of time; a separate petition accompanies this amendment.

I. Drawings

In response to the objection to the drawings under 37 CFR 1.83(a), applicant submits concurrently herewith a proposed drawing sheet of Figures 6A and 6B showing a thermal cyclers and optical system in accordance with embodiments of the invention and as clearly described in specification. Entry of this drawing sheet is respectfully requested.

II. Amendments

The specification has been amended to describe Figures 6A and 6B in the brief description of the drawings section and also to add reference numerals to components described in the detailed description section and shown in Figures 6A and 6B. No new matter has been added. Claims 23-29 have been canceled without prejudice, and new claims 30-47 have been added to more particularly emphasize various features of applicant's invention. Support for the claimed features recited in one or both of independent claims 30 and 39 is detailed below:

a reaction vessel, e.g., p.13, line 36; p.14, lines 3, 6, 11, 15, 18, 28;

an amplification reaction mixture comprising a target nucleic acid, reagents for nucleic acid amplification, and a

detectable nucleic acid binding agent, e.g., p.7, line 3 - p.8, line 8, p.9, lines 4-8;

contained in a sealed vessel condition, e.g., p. 6, lines 26-29; p. 8, lines 30-32; p.14, lines 24-26;

a thermal cyclor capable of alternately heating and cooling such a reaction vessel, e.g., p.9, lines 21-23; p.11, lines 20-22; p. 14, lines 9-23; p.22; Examples; and

an optical system including a detector operable to detect an optical signal related to the amount of amplified nucleic acid in the reaction vessel over a multiple-cycle period, with the reaction vessel in a sealed condition, allowing determination of a cycle-dependent change in such optical signal over a multiple-cycle period with the reaction vessel in its sealed condition, e.g., p.6, lines 22-23, 26-29; p.8, lines 30-31; p.13, lines 28-30; p.14, lines 1-34; p.20, lines 1-7; Examples; Fig. 5B.

Support for the dependent claims includes at least the following:

Claims 31 and 40, e.g., p.14, lines 10-23;

Claims 32 and 41, e.g., p.20, lines 1-7; Fig. 5B;

Claims 33 and 42, e.g., p.7, lines 7-10; p.16, lines 8-10;

Claims 34 and 43, e.g., p.7, line 35; p.19, line 17;

Claims 35 and 44, e.g., p.16, lines 26-27;

Claims 36 and 45, e.g., p.14, lines 14-34;

Claims 37 and 46, e.g., p.14, lines 14-34; and

Claims 38 and 47, e.g., p.21, lines 26-32; Examples.

### III. Rejections under 35 U.S.C. §103

The Examiner has rejected now-cancelled claims 23-29 under 35 U.S.C. §103 based on the 1989 Haff article entitled "Measurement of PCR Amplification by Fluorescence" in view of Mackay (EP 0 266 881). Although this rejection is moot in view of the cancellation

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of claims 23-29, applicant will explain how new claims 30-47 define over this art.

A. The Invention

The claimed invention is directed to an instrument and system for monitoring a nucleic acid amplification reaction over multiple thermal cycles. The instrument includes a thermal cycler capable of alternately heating and cooling, and adapted to receive, a reaction vessel containing an amplification reaction mixture comprising a target nucleic acid, reagents for nucleic acid amplification, and a detectable nucleic acid binding agent, in a sealed vessel condition. The instrument further includes and an optical system. The optical system includes a detector operable to detect an optical signal related to the amount of amplified nucleic acid in the reaction vessel over a multiple-cycle period, with the reaction vessel in a sealed condition, allowing determination of a cycle-dependent change in such optical signal over a multiple-cycle period with the reaction vessel in its sealed condition. An example of such a detected optical signal is illustrated in Figure 5B of the application.

B. The Cited Art

Haff is directed to an assay that measures DNA production by the fluorescent enhancement of a dye. A series of identical samples are prepared for amplification over multiple cycles. The samples are then successively withdrawn after different cycle numbers and the amount of DNA produced by the amplification process measured by fluorescence to yield a curve as shown in Figure 1 of Haff.

Mackay is directed to an optical assay system in which at least two target components of a sample mixture are labeled by respective fluorescent markers having different peak absorption or emission characteristics. There is no suggestion of (i) monitoring signal amplification in a sealed reaction vessel, (ii)

alternately heating and cooling a reaction vessel, or (iii) determining a cycle-dependent change in such a signal.

C. Analysis

An important difference between applicant's claimed instrument and the systems of Haff and Mackay lies in the sealed vessel condition under which amplification is carried out and optically detected. Neither Haff nor Mackay employ such a sealed vessel condition. In Haff's PCR amplification method samples are successively withdrawn after different cycle numbers and measured for fluorescence to generate a plot of DNA yield vs. cycle number. Applicant's claimed instrument is different. With applicant's invention, the amplification takes place and is detected in a sealed vessel condition. During amplification, a real-time signal indicative of a cycle-dependent change in double-stranded nucleic acid is generated to allow monitoring of the accumulation of double-stranded product while the amplification reaction is in progress, without opening the reaction vessel, without taking aliquots, and without withdrawing samples. Once the amplification reaction is initiated, no further handling or manipulative steps are required.

Nor are the deficiencies of the Haff reference remedied by Mackay. Mackay's system is directed to a different type of assay, the reference provides no motivation to modify the system to produce applicant's claimed invention. Also, unlike Mackay's system, applicant's claimed instrument does more than simply perform relative analysis; it allows target specific quantitation.

In addition to the structural and functional differences, applicant's claimed instrument offers significant advantages over the systems of Haff and Mackay. By carrying out amplification and detecting the optical signal under the sealed vessel condition, the reaction mixture cannot be disturbed or contaminated and the volume of the reaction mixture cannot be altered. Also, by

generating a signal that not only gives an indication of the inter-cycle net change of double-stranded product but intra-cycle variations as well, applicant's claimed instrument eliminates any ambiguities regarding the time of sampling. The signal generated by applicant's claimed invention is independent of the time of sampling.

Other features of applicant's invention are set forth in the newly added dependent claims.

Accordingly, it is respectfully submitted that neither Haff nor Mackay, whether taken individually or in combination, teach or suggest applicant's claimed instrument.

#### IV. Double Patenting Rejection

This provisional obviousness-type double patenting rejection is overcome in view of the cancellation of claims 23-29. Applicant further submits that new claims 30-47 are patentably distinct from the claims in copending application serial no. 08/266,061.

#### V. Conclusion

In view of the foregoing, applicant submits that the claims pending in the application comply with the requirements of 35 U.S.C. §112 and patentably define over the prior art. A Notice of Allowance is therefore respectfully requested.

If in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is encouraged to call the undersigned at (650) 324-0880.

Respectfully submitted,

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Date: 2/11/2000

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